Documented Outbreaks of Toxoplasmosis: Transmission of *Toxoplasma gondii* to Humans

JAMES L. SMITH

Eastern Regional Research Center, Agricultural Research Service, U.S. Department of Agriculture, Philadelphia, Pennsylvania 19118

ABSTRACT

Toxoplasmosis, a disease of mammals and birds, is caused by the obligate intracellular parasite, *Toxoplasma gondii*. Infection by the parasite is common (35-50% of the U.S. population are infected), but the disease, toxoplasmosis, is relatively rare and outbreaks of the disease are seldom seen. Documented outbreaks of toxoplasmosis are discussed to illustrate conditions that lead to outbreaks and how *T. gondii* is transmitted to humans. Recommendations are made of means to prevent introduction of the parasite into the environment and food supply as well as control mechanisms to prevent infection in humans and animals.

Infection by *Toxoplasma gondii* is generally silent with minor or no symptoms, but if the infected individual becomes immunocompromised, a serious case of toxoplasmosis may result. *Toxoplasma* is the cause of a common infection of humans in the United States; approximately half of the population is serologically positive to the parasite (*3*). Most, if not all of these individuals are immunocompetent and unaware of the infection. The disease, toxoplasmosis, is relatively rare. It is found mainly in immunocompromised individuals who comprise a small but steadily increasing segment of the population. Seronegative immunocompromised persons may be infected if they come in contact with *T. gondii*. However, most of the cases of toxoplasmosis in immunocompromised individuals represent recrudescence, i.e., reactivation of a previous infection. Thus, when seropositive individuals become immunocompromised, there is a good possibility that the parasite may be reactivated with resultant symptomatic toxoplasmosis (*56*). There is an increasing number of immunocompromised individuals in the U.S. population due to an increase in immunosuppressive therapies (organ and tissue transplants, treatment for cancer, kidney dialysis, etc.), to an increase in immunosuppressive diseases (AIDS, autoimmune diseases, etc.), and to an expanding population of less immunocompetent older people. There are an estimated 1,437,500 cases of *T. gondii* infection each year in the United States (*106*); most of these cases are probably asymptomatic. The number of deaths is estimated at approximately 300 with most of deaths due to congenital toxoplasmosis in neonates and toxoplasmosis in AIDS and other severely immunocompromised patients.

There are three infective stages in the life cycle of *T. gondii*: (i) sporozoites released from oocysts excreted during defecation by feline species, (ii) tachyzoites which are rapidly multiplying forms infecting animal cells, and (iii) bradyzoites encysted in various tissues and organs. Only in feline species do tachyzoites undergo sexual stages which result in the eventual formation of immature oocysts which mature (become infectious) and form sporozoites within the oocysts when feces from infected cats are exposed to the environment. The duration of oocyst excretion by the newly infected cat is 1 to 3 weeks and except in rare circumstances, the cat does not excrete oocysts again (*33*). When ingested, sporozoites are liberated from the oocysts in the intestinal tract and transform into tachyzoites which multiply in intestinal cells and then spread throughout the body of the host via the circulatory system. Tachyzoite multiplication is progressively inhibited by the developing immune response, and there is a shift to bradyzoite formation with eventual cyst formation in various tissues. Each cyst contains hundreds of bradyzoites. When meat containing tissue cysts is eaten, the cyst wall dissolves and the bradyzoites pass into the intestinal tract and are transformed into tachyzoites which infect cells of the new host with resultant parasitemia and eventual bradyzoite and tissue cyst formation. In immunocompromised individuals, the immune system can no longer keep the bradyzoites in tissue cysts and the bradyzoites are released and form tachyzoites (for a complete discussion of the life cycle of *T. gondii* see ref. 67).

How *T. gondii* infects man and animals was not understood for a long time. In a review on toxoplasmosis written in 1952, Weinman (*112*) stated:

> "The natural means of transmission (of toxoplasmosis) is unknown except in the case of congenital transmission."

However, in 1956, Weinman and Chandler (*113*) suggested that *T. gondii* could spread via a meat-to-man route and Jacobs et al. (*70*) demonstrated the presence of infectious *T. gondii* cysts in the diaphragms of sheep and swine. In a 1963 review, Jacobs (*68*) stated that the oral route, i.e., eating raw or undercooked meat containing *T. gondii* cysts, did indeed explain the incidence of toxoplasmosis in some populations and also explained how carnivorous animals become infected. However, Jacobs (*68*) believed that there must be

other routes of infection besides the congenital and meat eating routes since herbivorous animals and human populations that eat little or no meat may also show a high incidence of seropositivity to the parasite.

With the discovery of the infectious oocyst form of *T. gondii* in the feces of cats in 1970 (*59,66,98*), it was then possible to show that herbivorous animals and nonmeat eating people can be infected by *T. gondii* through inhalation of oocysts from dried cat feces or by ingesting foods contaminated with the feces of infected cats. Thus, there are three routes of infection: (i) environmental exposure to oocysts, (ii) carnivorous ingestion of bradyzoites in tissue cysts, and (iii) congenital exposure from tachyzoites crossing the placental barrier when a woman is infected during pregnancy. It appears that human toxoplasmosis has its origins in the use of animals either as a source of food or as pets.

OUTBREAKS

Outbreaks of toxoplasmosis are rarely seen since most infected immunocompetent individuals show few or no symptoms. Some documented incidents of *T. gondii* infection are discussed below with emphasis on the mode of transmission of the parasite.

On a British farm, four of six milking goats gave birth to weak or stillborn kids and the dams were shown to be seropositive to *T. gondii* (incident No. 1, Table 1). *T. gondii* cysts were isolated from one weak kid and milk from one doe was positive for tachyzoites. Domestic and feral cats were present in the immediate environment, and the goat feed (concentrates) was said to smell of cat urine. One member of the farm family, a son, developed mononucleosis-like symptoms during the investigation and was found to be seropositive. Another son was subsequently found to be positive for *Toxoplasma* antibody and had suffered a flu-like illness prior to the investigation. Both parents were seronegative. The members of the family shared several potential sources of infection: presence of cats, handling of goat feed and bedding that may have been contaminated by infected cat feces, gardening (cats may defecate in soft garden soil), eating raw vegetables from the garden, eating undercooked goat meat, and utilizing raw goat milk. The only differences between behavior of the parents and the sons was that the sons drank raw goat milk, whereas the parents only used goat milk in their hot tea or coffee (the hot liquids probably would destroy any tachyzoites present). It would appear that drinking raw goat milk led to *T. gondii* seroconversion in the sons in this family.

TABLE 1. *Documented outbreaks of toxoplasmosis.*

Incident No.	Probable route of infection	No. infected	Reference
1	Raw goat milk	2	*101*
2	Raw meat	4	*82*
3	Raw lamb	5	*100*
4a	Raw goat milk	2	*80*
4b	Raw goat milk	6	*80*
4c	Rare meat	2	*80*
4d	Rare meat	1	*80*
5	Raw or rare venison	3	*96*
6	Raw goat milk	10	*95*
7	Creek water	35	*5*
8	Geophagy (pica)	10	*103*
9	Inhalation	37	*105*
10	Rare lamb	6	*84*
11	Raw lamb	3	*53*
12	Raw goat milk	1	*89*
13	Raw beef	4	*10*
14	Geophagy (pica)	2	*54*
15	Raw or rare meat	1	*64*
16	Rare meat	95	*83*
17	Rare meat	5	*72*

In a 1-year study of 22 pregnant women in an Inuit village in northern Quebec, seven remained seronegative for *T. gondii* during pregnancy, 11 were seropositive before pregnancy, and four seroconverted (became infected) during pregnancy (incident No. 2, Table 1). None of the four newly infected women showed symptoms. As soon as their infections were diagnosed, the four women were placed on oral spiramycin therapy until delivery. Neonatal therapy for the babies at risk consisted of alternating courses of pyrimethamine/sulfadiazine and spiramycin. Two of the offspring appeared to have escaped infection, probably due to the early detection and treatment of their infected mothers. One child was probably infected and showed mild developmental delay. The other child showed clinical signs of toxoplasmosis at birth and developed cerebral calcifications, a ventricular cyst, and chorioretinitis.

Questionnaires administered to the 22 women indicated that seropositivity was significantly associated with skinning of animals for fur and with frequent consumption of caribou meat. Compared to seronegative women, the seropositives were >4 times more likely to have eaten dried seal meat, >6 times more likely to have eaten seal liver, and >8 times more likely to have eaten raw caribou meat. Cats are not kept by the Inuits and were not correlated with infection in these women. Guidelines for prevention of *T. gondii* infection in seronegative pregnant women in the arctic region should include not eating uncooked or dried meats (particularly seal and caribou) and avoiding the skinning of animals.

Serology of an Australian child infected with hepatitis A also indicated that the child had a high antibody titer for *Toxoplasma* (incident No. 3, Table 1). Investigation of the remaining family members indicated that three other children and the mother had high antibody titers to *T. gondii*, but the father was seronegative. The authors suggested that this family outbreak was due to the eating of raw lamb. The family was Lebanese in origin and often served Kibbi, which is prepared from lamb and ground cracked wheat. The dish is sometimes fried but also can be eaten raw.

In incidents No. 4a-4d (Table 1), studies had been initiated to determine the frequency of acute *T. gondii* infections in family members of an index patient with acute lymphadenopathic toxoplasmosis. Incidents No. 4a (Washington state) and No. 4b (California) were probably caused by ingestion of raw goat milk, and incidents No. 4c and No. 4d (both California) were probably from eating undercooked meat. Five other incidents were discussed by the authors, but the source of infection was not determined. The results obtained by Luft and Remington (*80*) suggest that when an index patient has lymphadenopathic toxoplasmosis, other family members may show that they too have been infected by *T. gondii*.

In incident No. 5 (Table 1), three deer hunters became acutely ill and required hospitalization after consuming raw or grossly undercooked venison. The individuals presented with fever, leukopenia, and abnormal liver function as well as other symptoms and were seropositive for *T. gondii* antibody. All three individuals admitted to liking venison either raw or very lightly cooked. Since two of the individuals involved were state wildlife workers, the authors examined a group of South Carolina and Georgia wildlife workers who handled deer viscera on a regular basis and found that 19 of 62 individuals (30.6%) were seropositive. Also, there was a significant association between toxoplasma seropositivity and eating raw or rare venison among these workers.

In incident No. 6 (Table 1), a woman who lived on a ranch in northern California had fever, tiredness, and headaches. One month later, retinochoroiditis was diagnosed and serology demonstrated a high titer for *T. gondii* antibodies. The patient's condition improved after treatment with pyrimethamine, sulfadiazine, prednisone, and folinic acid. Examination of 24 family members indicated that

10 members were seropositive (including the index case). There were a number of animals on the ranch including horses, swine, ducks, sheep, dogs, cats, and goats. Milk was supplied by three milking goats, two of which were seropositive for *Toxoplasma* (the third goat had been sold and could not be located). A cat and a goat kid were seropositive also. None of the other animals on the ranch were positive for *T. gondii*. All 10 of the seropositive individuals drank raw goat milk, whereas the 14 seronegatives did not.

In incident No. 7 (Table 1), 600 U.S. soldiers were involved in jungle training for 2-3 weeks in the Panama Canal zone area. After their return to the United States, 32 of the soldiers developed various symptoms including fever, chills, headache, and malaise. Serological investigation indicated that these individuals (plus three others who were asymptomatic) had high antibody titers to *T. gondii*. An epidemiological investigation indicated that positive serology was correlated with consuming water from jungle creeks. It is probable that the *T. gondii* oocyst-containing water had been contaminated by feces from infected jungle cats.

Ten individuals (mostly preschoolers) from an extended family of 30 living in southern United States showed evidence of *T. gondii* seroconversion during incident No. 8 (Table 1). The index case, a 3 1/2-year-old boy presented with severe symptoms of toxoplasmosis including brain abscesses, progressive chorioretinitis, seizures, neurological deficits, hepatosplenomegaly, pneumonitis, and eosinophilia. Toxoplasmosis was confirmed by serology and identification of the organism in the brain and cerebrospinal fluid. This child also was infected by *Toxocara*. Five other children (<5 years of age) and one adult showed symptoms of acute toxoplasmosis and positive serology. Two teenagers and one adult seroconverted during the investigation but remained asymptomatic. Serology indicated that six other family members had been infected by *T. gondii* in the past; a total of 16 family members were seropositive. Cats were present nearby and were allowed to defecate in the area where the children played. An epidemiological study indicated that the childrens' play area was a 'natural sandbox' (a moist, loosely packed, sandy surface). Of the 11 children ≤5 years old, eight had a history of geophagia and seven of these were seropositive for *T. gondii*; four of the 11 were positive for *Toxocara*. It is probable that the children (including the index case) were infected by ingesting oocyst-containing soil from the playing area. It is not certain how the remaining older individuals were infected but undercooked meat was not implicated. These individuals may have been infected by inhalation of oocysts from dust of the children's play area. The index child presented with more severe symptoms than normally would be expected; his toxoplasmosis was not congenital because his mother was suffering from acute toxoplasmosis during the investigative period. The authors suggested that the child's severe clinical manifestations resulted from simultaneous infection with *Toxoplasma* and *Toxocara*.

When a patient presented to a physician with symptoms of toxoplasmosis, she told him that she knew of other people with similar symptoms (incident # 9, Table 1). An investigation indicated that 37 patrons of a local riding stable in the Atlanta area had high antibody titers against *T. gondii*; 35 of these individuals were symptomatic with >80% suffering from fever, headache, and lymphadenopathy. One seropositive woman patron who was pregnant (first trimester) elected to have an abortion; *T. gondii* was isolated from fetal material. *T. gondii* was also isolated from trapped mice and feral cats and kittens. The floor of the indoor arena of the riding stable was silt, and cats were known to have defecated in the arena area. Epidemiological studies indicated fecal oocysts were probably inhaled by the infected patrons from dust stirred up by horses or from dragging and smoothing operations of the arena or from ingestion of food or beverages contaminated by oocyst-laden dust. The unusual aspects of this outbreak was the large number of symptomatic individuals. Dubey et al. (*46*) studied the *T. gondii* isolated during the outbreak and found that the oocysts from these isolates were more pathogenic for mice than were the tachyzoites or cysts of these isolates. Similar heightened pathogenicity of oocysts might be expected in humans infections.

Six of seven household members in New York state demonstrated high *T. gondii* antibody titers consistent with recent infection (incident No. 10, Table 1). Five of the people were symptomatic. The most common clinical manifestations were fever and lymphadenopathy. One adult (the index case) developed retinochoroiditis which was treated with prednisone, sulfadiazine, pyrimethamine, and folinic acid; however, his vision did not improve. The source of infection was not identified, but ingestion of rare lamb was considered as the most probable source.

Two patients suffered from severe frontal headache, fever, lymphadenopathy, and malaise, but a third patient was asymptomatic in a London toxoplasmosis outbreak believed to be due to the ingestion of inadequately grilled lamb (incident No. 11, Table 1).

At the Sacramento Medical Center, an underweight, 7-month-old male presented with failure to thrive, vomiting, dehydration, lymphadenopathy, sore tongue and mouth, and fever (incident No. 12, Table 1). The infant had been breast-fed until two months of age and then he was switched to raw goat milk. Serology indicated that the child did not have brucellosis or Q fever but did have antibodies for *Toxoplasma*. Treatment with pyrimethamine and trisulfapyrimidines led to weight gain and increased fluid intake. Serology of 10 goats in herds that furnished the baby's milk indicated that four of them were seropositive for *T. gondii*, but tachyzoites were not found in milk samples. All 10 of the goats were seropositive for *Coxiella burnetii* but not *Brucella*. Epidemiological evidence indicated that the child was infected from drinking raw goat milk. While tachyzoites are rapidly killed by stomach conditions, they could have penetrated lesions present in the buccal mucosa. The child's mother was seronegative; therefore, congenital toxoplasmosis was ruled out.

Initially, two male members of a wedding party in Pennsylvania presented with low-grade fever, chills, generalized fatigue, and swollen cervical lymph nodes in incident No. 13 (Table 1). A few weeks later, another male member showed similar symptoms while a female member had similar symptoms but no lymphadenopathy. All showed high *T. gondii* antibody titers indicative of recent infection. These four people were attendees at a 19-member wedding party supper held at a Syrian restaurant. Fifteen members of the party, including the four ill individuals, admitted eating Kibee Nayee, a meat dish made from raw beef. It appears likely that eating the raw meat dish led to this toxoplasmosis outbreak. Of the 15 members contacted (including the ill individuals), nine (60%) were seropositive. Of the nine seropositive individuals, six (66.7%) had a habit of eating raw or rare meat. Six of the 15 (40%) were seronegative, and only one of these six individuals habitually ate raw or rare meat. These data, while limited, suggest that seropositivity to *T. gondii* was correlated with raw or rare meat consumption.

The index case in incident No. 14 (Table 1), a woman, became ill with lymphadenopathy and demonstrated high titer of *T. gondii* antibody; a few months later, one of her children, a son, demonstrated a high titer. Oocysts were present in a sand pit in the garden but not in garden soil. A neighbor's cat, which was seropositive, had used the sandpit to deposit its feces. The son was probably infected when playing in the sandpit, but it is not clear just how the mother came in contact with the oocysts.

Adult acquired *Toxoplasma* encephalitis was diagnosed in an operating room nurse from a London hospital (incident No. 15, Table 1). Her history indicated that she habitually ate rare and occasionally, raw steak; her husband did not eat raw or rare meats and was seronegative. Treatment with pyrimethamine, sulfadiazine, and folinic acid was successful in relieving the woman's symptoms.

An epidemic of toxoplasmosis occurred at a university in Sao-Jose-dos-Campos, S. P., Brazil, which involved 95 students (incident No. 16, Table 1). Seventy-seven students were symptomatic,

whereas 18 were not, but all had high antibody titers against *T. gondii*. Most of the students said that they had eaten undercooked meat at the college cafeteria. However, the source of the outbreak was not determined with certainty.

In incident No. 17 (Table 1), the index case, a male medical student at Cornell Medical College, presented with fever, myalgia, headache, and lymphadenopathy and demonstrated a high titer to *T. gondii*. Three male and one female students were also symptomatic and had high titers. All five students had eaten rare hamburgers served at a campus snack bar on the same occasion. All of the ill students attended different classes, lived at different locations, ate separately, and were not well-acquainted. The only thing that they appeared to have in common was eating rare hamburger on that one occasion. The snack bar operator denied mixing the beef with either mutton or pork and asserted that the meat grinder had been used only for beef. Evidence indicated that the source of infection was the rare beef hamburgers.

DISCUSSION

Documented outbreaks suggest that oocysts, if they are inhaled or ingested, can cause *T. gondii* infection. Since the oocysts are relatively resistant to gastric juices (more so than either tachyzoites or bradyzoites), they survive passage through the gut (*38*). Oocysts in feces are stable to drying: at relative humidities ranging from 22 to 80%, infectivity persisted for 14-18 d. Oocysts are stable in water at 20-22°C for more than a year and can survive for at least a year in soils in either tropical or temperate climates (*33,57,94*). Oocysts, inoculated in soils in Kansas and Costa Rica, survived for 12-18 months (*60*) which indicates that oocysts can survive hot summers and cold winters. Yilmaz and Hopkins (*116*) demonstrated that covered cat fecal deposits retained infectivity for 153-410 d at temperatures ranging from 4-47°C. Oocysts in uncovered feces were viable for 30-214 d. Therefore, the stability of oocysts to environmental conditions ensure that they can infect anyone who comes in contact with them.

That oocysts are present in soils indicate that they may be found in coprophagous insects or earth worms. Frenkel et al. (*60*) isolated infectious oocysts from earth worms and Wallace (*109,110*), and Chinchilla and Ruiz (*11*) recovered oocysts from cockroach feces and body surfaces of flies. These insects, as well as earth worms, can perpetuate *Toxoplasma* by serving as food for birds and rodents. Insects also may possibly transport oocysts to foods.

Remington et al. (*88*) and Frenkel and Ruiz (*58*) demonstrated that children in Central America seroconvert to *Toxoplasma* early so that approximately 60% are seropositive by age 20. In North America, however, only 15-20% are seropositive at age 20. The authors postulate that the seropositivity pattern seen for Central America (and probably Latin America as a whole) is due to oocyst infection while that of North America is due to infection from eating contaminated meats. Because of low levels of hygiene and the abundance of feral cats, children in Central and Latin America come in contact with fecal oocysts while playing in areas where the soil or surfaces have been contaminated by feral cat feces. Similar conclusions were reached by Wallace (*111*) for the incidence in many of the Pacific Islands; by Ghorbani et al. (*62*) for northern Iran; by Barbier et al. (*4*) for La Guadeloupe, French West Indies; and by Sousa et al. (*102*) for Panama. Seroconversion in those nonindustrialized areas appears to be due to infection by *T. gondii* oocysts and not to meat eating.

Working with oocysts in laboratory settings also has resulted in seroconversion. Miller et al. (*85*) described the seroconversion of seven laboratory personnel who had worked for an average period of 10 years with tachyzoites and cysts without seroconversion, but serocoversion occurred after they had worked with oocysts for an average of 2.3 years. Thus, *T. gondii* oocysts can be infectious to individuals who come in contact with them.

Raw goat milk has been implicated as a source of *Toxoplasma* infection in documented outbreaks; the infectious form would be tachyzoites. Tachyzoites are the least environmentally stable form of *T. gondii*. Exposure to 50°C renders them non-infectious, and therefore, normal milk pasteurization temperatures will eliminate tachyzoites (*38*). Nor are tachyzoites stable to artificial gastric juice (pepsin and trypsin); survival time was less than 60 min (*71,97*). However, there may be survival of tachyzoites if gastric juice is low in acidity (*71*). Thus, tachyzoites would not normally be expected to survive passage through the gut.

T. gondii was detected in milk from experimentally infected goats but not from naturally infected goats (*16,48*). Since tachyzoites are destroyed by gastric juice, it is probable that the risk of infection from drinking raw goat milk is low. However, the presence of lesions in the oropharyngeal area (due to poor nutrition, vitamin C deficiency, or to cuts from brushing the teeth) may allow tachyzoites to penetrate the mucosa. Conditions under which stomach acids may be neutralized—type of food, presence of fat, or drugs—may allow tachyzoites to escape to the more alkaline duodenum unharmed (*95*). However, proper pasteurization would eliminate *T. gondii* infection from drinking goat milk.

Eating raw or undercooked meats was the probable cause of *Toxoplasma* infection in most documented outbreaks (Table 1); a number of epidemiological studies bear this out. French children who had long stays in tuberculosis hospitals showed high seroconversion rates to *T. gondii*. At the Leon Bernard Hospital, 30% of previously seronegative children seroconverted during their hospital stay (*14*). The child patients of this hospital were fed raw minced meat twice weekly. Out of 1,125 children admitted to the tuberculosis section of the Brevannes Hospital, 641 were *T. gondii* seronegative at the time of admission but 204 seroconverted during their hospital stay. It was part of the therapeutic regimen of the hospital to feed the children raw or underdone meats (*15*). At both of these hospitals, it appeared that the serving of raw or undercooked beef, horse, lamb, or mutton contributed to the observed increases in *Toxoplasma* seropositivity.

Knaus (*73*), studying the relationship between raw meat eating and *T. gondii* seropositivity in German pregnant women, found that 2,205 out of 5,012 (43.9%) pregnant women were seropositive and admitted to eating raw meat. However, only 319 out of 1,049 (29.2%) pregnant women who did not admit to eating raw meat were seropositive for *Toxoplasma*. Braveny et al. (*8*) studied 337 German patients with acute toxoplasmosis and found that 42% were raw meat eaters, and 19.9% had close contact with cats. Only 21% of 198 seronegative controls admitted to eating raw meat, and only 6.7% had close cat contact. In a study of a farming community in Miki

City, Japan, Konishi and Takahashi (*74*) discovered that 221 of 975 men (28.8%) were seropositive. Seropositivity increased from 1.6% at 20-29 years of age to 47.2% by 70-90 years of age. Of 2,631 Japanese women, 429 were seropositive (16.3%) which increased from 3.6 at age 20-29 to 32.9% by ages 70-90. Konishi and Takahashi (*74*) reported that 38.9% of the men and 34.6% of the women in Miki City had contact with cats; however, 41.6% of the men were raw meat eaters in contrast to 17.2% of the women. Beef, chicken, horse, whale, wild boar, and venison were the sources of raw meat. The picture is quite different for Guangdong in the Peoples Republic of China where the human prevalence for *T. gondii* antibody is low (<1%). Shen et al. (*99*) attributed the low level of *Toxoplasma* infection to the Chinese habit of cooking meat well done and to the rarity of cats in the home environment. Thus, there does appear to be a strong association with *Toxoplasma* seropositivity and raw or lightly cooked meat consumption.

The presence of *T. gondii* cysts in edible meats is well documented (Tables 2-3). Bradyzoites, which are found in tissue cysts, are more resistant than tachyzoites to the acid conditions of the stomach and can survive in artificial gastric juice for at least 2 h. Thus, they will survive the stomach environment long enough to infect the animal or human who eats meats containing tissue cysts (*71,97*).

The data presented in Table 2 indicate that the favorite game of most hunters in the United States, deer, moose, and elk, can harbor cysts of *T. gondii* in edible tissue. However, information on the extent of *T. gondii* infection in wild game is scanty. Franti et al. (*55*) reported that 77 out of 382 (20%) Columbia black-tailed deer in northern California were seropositive to the parasite. Only one of 30 white-tailed deer in Florida was positive for *T. gondii* antibody (*9*); however, Lindsay et al. (*79*) found seven out of 16 white-tailed deer in Alabama were seropositive. Dubey (*25*) tested 56 elk in Montana, and all were seronegative. There is no doubt that animals in the wild can be and are infected by *T. gondii* under natural conditions. Pronghorns and mule deer, intraruminally inoculated with *T. gondii* oocysts, showed clinical signs of illness and died within a short period. At necropsy, typical lesions of acute toxoplasmosis were present (*41*). Experimentally infected elk were asymptomatic, but 73 d after infection, cysts were present in various tissues (*40*). Most of the wild game animals hunted by man are not carnivores and are probably infected by ingesting or inhaling oocysts from feces deposited by feral or wild cats. Small urban wild game animals, such as squirrels and rabbits, probably are infected when they come in contact with oocysts from the feces of domesticated cats. Wild game animals obviously represent reservoirs of *T. gondii* (Table 2) and can infect humans who hunt and eat them. Viscera from animals that are field dressed should be burned or buried to prevent infection of wild felines, carnivores, and omnivores.

T. gondii cysts are found in various tissues, including edible tissue, of fowl (Table 3). Chickens infected with *T. gondii* do not usually show clinical signs of disease. Laying hens inoculated directly into the crop with 5,000 *T. gondii* oocysts did not show clinical signs even though cysts were present in brain and heart at 6 weeks after infection (*6*). When hens received 50,000 oocysts, there was a decrease in egg production but no other sign of illness; *T. gondii* cysts were present in heart and brain tissues of these hens 6 weeks after infection. Older methods of serology for toxoplasmosis (in particular, the Sabin-Feldman dye-test) often give negative results when used with fowl; however, Biancifiori et al. (*6*) by using enzyme-linked immunosorbent assay (ELISA), showed that all hens were seropositive by the second week after infection. Jacobs and Melton (*69*) recovered *T. gondii* from one of 327 eggs laid by infected hens; however, Biancifiori et al. (*6*) did not find the parasite in any of 550 eggs laid by infected hens. Pigeons, inoculated via the crop with 50 oocysts, did not show clinical illness but did demonstrate seropositivity (ELISA), and *T. gondii* cysts were present in various tissues at 45 d after infection. Birds receiving 500-5,000 oocysts showed clinical signs of illness which eventually led to death (*6*). Thus, *T. gondii* cysts persist for some weeks in chickens and pigeons (and probably other poultry), but since poultry is normally eaten well done, it is probable that eating poultry is not a common source of infection. While extensive studies have not been done, eggs appear to be a negligible source in transmission of *T. gondii*.

Cysts of *T. gondii* have been demonstrated in edible tissues of horses, ponies, and mules (Table 3). In a nationwide study, Riemann et al. (*91*) found that 20% of 1,294 U.S.

TABLE 2. *Wild game in which* T. gondii *cysts have been demonstrated in one or more tissues by use of the cat and/or mouse test.*[a]

Animal	Type of infection	Tissue where cyst found	Reference
Moose	Natural	Muscle	*18*
Pronghorn	Natural	Muscle	*18*
	Experimental	Brain, diaphragm, heart, lung, semitendinosus muscle	*41*
Mule deer	Natural	Muscle	*20*
	Experimental	Brain, diaphragm, heart, kidney, liver, semitendinosus muscle	*41*
Elk	Experimental	Brain, diaphragm, heart, liver, pancreas, skeletal muscle	*40*
Roe deer	Natural	Diaphragm, esophagus, heart muscle	*50*
White-tailed deer	Natural	Heart	*79*
Farmed red deer	Natural	Diaphragm, brain	*12,114*
Gray squirrel	Natural	Brain, heart, lung, skeletal muscle	*92*
Rabbit	Experimental	Brain, heart, kidney, liver, lung, spleen	*85*

[a] Mouse assay: suspect cyst-containing tissue is homogenized, pepsinized, and inoculated into mice: 6-8 weeks later, brains of mice are examined microscopically for *T. gondii* cysts. Cat assay: suspect cyst-containing tissue is fed to cats; within 2 weeks, infected cats should excrete oocysts which are detected microscopically in cat feces and/or by feeding fecal oocysts to mice (*33*).

TABLE 3. *Domesticated animals in which* T. gondii *cysts have been demonstrated in one or more tissues by use of the cat and/or mouse assay.*[a]

Animal	Type of infection	Tissue where cyst found	Reference
Chicken	Natural	Ovary, oviduct	*69*
	Natural	Brain, heart	*17*
	Experimental	Brain, breast & leg muscle, gizzard, kidney, ovary, oviduct	*69*
	Experimental	Brain, heart, liver, lung, spleen	*85*
	Experimental	Brain, heart, liver, lung	*6*
Japanese quail	Experimental	Brain, heart, kidney, liver, lung, skeletal muscle, spleen	*85*
Pigeon	Experimental	Brain, heart, kidney, liver, lung, muscles, spleen	*6*
Horse	Natural	Diaphragm, heart	*1*
Pony	Experimental	Brain, diaphragm, heart, kidney, liver, skeletal muscle	*2*
Horse, mule, pony	Experimental	Brain, heart, lung, muscle, tongue	*24*
Calves	Experimental	Diaphragm, lung, muscle, spleen	*13*
	Experimental	Pool of various tissue including skeletal muscle	*52*
	Experimental	Brain, diaphragm, kidney, liver, lung, skeletal muscle, tongue	*22*
Pregnant cows	Experimental	Brain, liver	*104*
	Experimental	Heart, liver, skeletal muscle	*22*
Kids	Transplacental	Brain, diaphragm, heart, kidney, liver, lung, skeletal muscle, spleen	*17,19*
Does	Natural	Diaphragm, heart, kidney, liver, thigh muscle	*17*
	Experimental	Brain, diaphragm, heart, kidney, liver, thigh muscle	*21*
Adult goats	Natural	Brain, diaphragm, heart, kidney, liver, skeletal muscle	*16*
	Experimental	Brain, diaphragm, heart, kidney, liver, skeletal muscle	*16*
	Experimental	Brain, diaphragm, heart, kidney, liver, lung, pancreas, skeletal muscle, spleen	*48*
Sheep	Natural	Diaphragm	*108,115*
	Experimental	Brain, diaphragm, heart, kidney, liver, lung, pancreas, skeletal muscle, spleen	*36*
	Experimental	Brain, diaphragm, heart, kidney, liver, thigh muscle	*23*
Lamb	Congenital	Heart, intercostal and limb muscles, tongue	*34*
	Experimental	Brain, liver, lung, muscle	*23*
Pig	Natural	Diaphragm	*115*
	Natural	Diaphragm, heart, skeletal muscle, tongue	*43*
	Experimental	Muscle	*49*
	Experimental	Brain, diaphragm, heart, kidney, liver, thigh muscle, tongue	*39*
	Experimental	Brain, diaphragm, heart, liver, longissimus muscle	*87*
	Experimental	Brain, diaphragm, heart, kidney, skeletal muscle, tail, tongue	*43*
	Experimental	Brain, diaphragm, heart, skeletal muscle, tongue	*28*

[a]See footnote, Table 2.

horses were seropositive for *T. gondii*. Antibody to the parasite was present in 10% of 500 horses slaughtered at a horse meat packing plant (*1*), and pooled tissues from the horses contained cysts. Equids appeared to be quite resistant to clinical toxoplasmosis, but *T. gondii* cysts persisted in tissue for at least 476 d (*24*). Since horses are herbivores, they are probably infected by ingesting or inhaling oocysts present in feeds, hay, and bedding contaminated by cat feces.

Calves and cows, experimentally infected with *T. gondii*, showed evidence of tissue cysts in various tissues (Table 3). Dubey and Streitel (*37*), utilizing heart and diaphragm tissue from 352 cattle slaughtered at an Ohio abattoir, found no evidence of toxoplasmosis, suggesting that market cattle may have a low incidence of *Toxoplasma* infection. Calves, 3 months of age, inoculated with oocysts (10,000-100,000 orally) or tissue cysts (1,000 subcutaneously) developed clinical signs of illness and parasitemia, and cysts were present in various tissues for at least 107 d after infection (*13*). Fayer and Frenkel (*52*) demonstrated clinical signs of illness and presence of *T. gondii* cysts in tissue of 3-month-old calves which had received oral doses of 100-1,000 oocysts; cysts persisted in tissues for up to 90 d after infection. In a review of toxoplasmosis of cattle, Dubey (*26*) presented a table summarizing attempts, worldwide, to isolate *T. gondii* from tissues of naturally infected cattle. Out of 4,302 infected cattle, 201 (approximately 5%) were positive for *T. gondii* cysts in one or more tissue. Thus, there appears to be a low prevalence of *T. gondii* in cattle. The serologic prevalence of *Toxoplasma* in cattle is unknown since most of the serological tests give nonspecific results with bovine sera and estimates of disease in cattle are too high (*26,29*). Since serology is not reliable, polymerase chain reaction may be a technique that can be used to give better estimates of the prevalence of *Toxoplasma* in cattle. Such a procedure has been used for

humans (*107*) but does not appear to have been used with cattle. Dubey (*26,29*) made some conclusions concerning the role of toxoplasmosis in cattle: (i) *T. gondii* does not appear to be important in causing clinical illness and/or abortions, but few studies have been done; (ii) *T. gondii* appears to be quickly eliminated from bovine tissue; and (iii) milk from infected cows does not appear to be implicated in transmission of the parasite. Since the status of *T. gondii* and toxoplasmosis is uncertain in cattle, the role of beef in transmission of the disease to humans cannot be adequately evaluated at the present time.

T. gondii induces a more severe disease in goats than in other domestic animals and clinical signs are often seen. A *T. gondii* infection during pregnancy can cause early embryonic death and resorption of the fetus, fetal death, mummification, abortion, still birth, or neonatal death. Surviving kids may be infected with the parasite (*31,45*). Abortion and congenital toxoplasmosis in goats can lead to severe economic loss, especially to the small dairy goat farmer. Losses from toxoplasmic abortion can be minimized by retaining, as breeding stock, does that abort because they generally will not abort again (*17*). Of 2,334 goats tested in various part of the United States, 23% were seropositive for *T. gondii* (*31,32*). Seropositivity increased with age: 6-month-old goats had a seropositivity of 3.7%, whereas 34.5% of goats 5-10 years old were positive (*32*). Cysts are found in various tissues of goats that have been naturally, transplacentally, or experimentally infected with oocysts (Table 3). The cysts persist in transplacentally infected kids for up to 235 d after birth (*19*) and persist in the livers, kidneys, and other edible tissue of experimentally infected adult goats for as long as 441 d after infection (*21*). The persistence of the parasite in goat tissue indicates that goat meat can be an important source of *T. gondii* to humans, especially to members of ethnic groups in which goat meat is an important part of the diet. These individuals will probably butcher the goat themselves and should be aware that cutting through tissue cysts can release bradyzoites which can cause an infection by entering a wound. Other foods can be contaminated via hands or contaminated equipment unless thorough cleansing is done. Goats are probably infected by ingestion or inhalation of oocysts present in feeds, hay, or bedding contaminated by the feces of barn cats.

When *T. gondii* infects sheep, clinical symptoms are generally not seen; however, in pregnant sheep, infection can result in early embryonic death, abortion, still birth, or neonatal death (*86*). Abortion occurring in 1,757 sheep investigated by two U.S. diagnostic laboratories indicated that 16.3% of the abortions were due to *T. gondii* infection (*31*). Ewes that abort once from *T. gondii* infection do not usually abort again, and thus, the sheep raiser should not cull these animals from the flock but use them for breeding stock (*35,90*). The seroprevalence of *T. gondii* in 4,880 sheep from various parts of the United States was 42.9%, whereas the seroprevalence in 1,056 lambs was 8% (*31*). *T. gondii* cysts are present in a number of tissues in lambs and sheep (Table 3). In 7-month-old congenitally infected lambs, cysts were present in tongue, lamb chops, and leg of lamb (*34*). In experimentally infected sheep, *T. gondii* cysts were found in skeletal muscle at 119 d after infection (*36*). Thus, it appears that *T. gondii* cysts persist in sheep and lambs for long periods. Toxoplasma infection in sheep appears to be sporadic and is due to inhalation or ingestion of oocysts from contaminated hay, bedding, or feed. Faull et al. (*51*) have indicated that infection of sheep may occur in pastures on which oocyst-contaminated bedding has been spread; grazing on such pastures can expose the animals to oocysts.

Most *T. gondii* infections in swine are subclinical with clinical toxoplasmosis occurring most commonly in neonates and young pigs. Transplacental infections in pigs are less common than are postnatal infections, but congenital toxoplasmosis is seen in young delivered from infected pregnant sows (*27,30*). In a recent national survey for *T. gondii* seroprevalence in U.S. swine, Dubey et al. (*47*) found that 24% of 11,842 pigs were seropositive. Dubey (*27*) summarized, in table form, the literature on the *Toxoplasma* seroprevalence in swine from various countries. Assuming that the different serological assays used are equivalent, the seroprevalence for the parasite worldwide in swine is approximately 22% of 737,117 pigs surveyed. Cysts of *T. gondii* have been detected in several edible tissues of pigs (Table 3). Cysts were present in the tissue of pigs at 171 d (*39*), 357 d (*43*), and 875 d (*28*) after infection. Thus, tissue cysts persist in swine well beyond the life of pigs raised for market. Dubey (*28*) and Dubey et al. (*43*) demonstrated that tissue cysts persist in pig muscles used for commercial pork cuts such as the arm picnic, ham, Boston butt, spareribs, tenderloin, bacon, and tailbone. Rothe et al. (*93*) sampled pork chops from 15 butcher shops in metropolitan Adelaide (Australia); only one chop out of 30 contained *T. gondii* cysts. The omnivorous habits of swine greatly enhances their contact with *T. gondii*. Precautions that can control infections in swine include cooking all garbage to be fed to swine, not allowing cats to contaminate grains and ground feeds with feces, and not allowing cats around hog-raising facilities (garbage feeding leads to rodents which should be controlled by means other than cats). Pigs are cannibalistic, and precautions are needed to prevent tail biting and eating of dead or weakened pigs. It is probable that the rooting nature of pigs exposes them to oocyst-containing soils, and therefore, exclusion of cats from swine-raising facilities is a must.

It is obvious that meat animals must be protected against *T. gondii* infection. Therefore, cats should not be allowed in livestock housing or feeding areas. Cats are known to defecate in ground animal feeds, hay, and bedding. If the cats are excreting *T. gondii* oocysts, other animals can be infected. Nor should cats be used for rodent control in animal-raising facilities.

Since it is possible that an animal carcass may contain *T. gondii* cysts, individuals who handle raw meats should exercise caution if they have wounds on their hands or arms because rupture of tissue cysts by cutting or grinding will release bradyzoites that can penetrate wound sites. Hands and all butchering tools, equipment, and tables should be thoroughly cleansed to prevent cross-contamination. During preparation of meat dishes, raw meat should not be tasted and raw meats should not be fed to pets; in particular, raw meats should never be fed to cats.

Dubey et al. (*44*) determined the D values for the heat destruction of tissue cysts present in homogenized pork

muscle. The D values at 49, 55, 61, and 67°C were 22.0, 5.1, 3.7, and 3.6 min, respectively. The D values for inactivation of *Trichinella spiralis* larvae in ground pork muscle at 49, 52, 55, and 60°C are 557, 60.5, 7.0, and <2 min, respectively (*75*). Thus, *T. spiralis* are more heat resistant than *T. gondii* and a trichinae cook would inactivate *Toxoplasma* cysts present in meats. Similarly, *T. gondii* cysts are less resistant than *T. spiralis* to freezing. Complete and instant inactivation of *Toxoplasma* cysts in ground pork muscle occurs at -12.4°C (*76*), whereas -23.3°C was needed to ensure inactivation of *Trichinella* larvae in ground pork muscle (*77*). Thus, temperature treatments of meat (heating and freezing) that inactivate *T. spiralis* will inactivate *T. gondii*, also. Lunden and Uggla (*81*) investigated the effect of microwave cooking on destruction of encysted *T. gondii* in mutton. Cooking the meat to an internal temperature of 65°C did not always ensure inactivation of the parasite. They noted that portions of the meat cuts were reddish in color, which indicated that microwave heating was uneven and resulted in undercooking.

Using either a cesium-137 or cobalt-60 source, Dubey et al. (*42*) were unable to detect infectious *T. gondii* cysts in ground pork muscle after a 50-Krad treatment. But, Brake et al. (*7*) found that treatment of ground pork muscle containing *T. spiralis* larvae with 15-20 Krad from a cesium-137 source led to inactivation of the larvae. Brake et al. (*7*) believed that treatment of pork with 30 Krad would provide an adequate margin of safety for irradiation destruction of *Trichinella*. Since *T. gondii* is more resistant than *T. spiralis* to radiation, 75-100 Krad would be necessary to ensure destruction of *Toxoplasma* infectivity in pork and other meats.

It is generally assumed that meat curing conditions which inactivate *T. spiralis* will inactivate *T. gondii*, also. However, essentially no data exist for the effects of meat curing agents (NaCl, nitrite, ascorbate, etc.) or conditions (drying, fermentation, etc.) on *Toxoplasma* cysts.

CONCLUSION

T. gondii as tissue cysts may persist for the life of the individual. At the present time, there is no definite cure for toxoplasmosis because no drugs that will kill bradyzoites present in tissue cysts are available (*61,65*). Therefore, prevention of infection is the only effective means of toxoplasmosis control. Raw goat milk, apparently an important source of *T. gondii* infection (Table 1), can be pasteurized. The information in Table 1 also indicates that meat is a major means of transmitting *T. gondii* infection, and the elimination of raw and/or rare meats from the diet by thorough cooking (internal temperature of 70°C) of meat will prevent a large number of infections. Alternatively, freezing (lower than -12.5°C) or irradiating (75-100 Krad) meats will render bradyzoites noninfective.

However, the presence of oocysts in the environment would still pose a threat, even after using all of the preventive measures listed above. Elimination of cats from livestock-raising facilities will prevent the deposition of cat feces in feeds, hay, and bedding. Preventing house cats or kittens from hunting will ensure that they will not be infected from rodents or birds. Alternatively, the prospective cat owner should buy only *T. gondii*-seropositive cats for pets because they will not excrete oocysts again (except in cases when the cat may be immunocompromised). Since oocysts need 24-48 h of environmental exposure to become infectious after defecation by an infected cat, daily disposal of cat litter should prevent exposure of the owner to mature and infectious oocysts.

The logical approach to control of toxoplasmosis would appear to be vaccination. Hermentin and Aspock (*63*) and Leighty (*78*) discuss various aspects of vaccines against *T. gondii*. A vaccine would be useful in protecting domestic cats against infection and would greatly reduce oocyst contamination of the environment. However, vaccination of cats has its limits since there are a large number of feral and stray *Felis catus* in the environment. A vaccine against *T. gondii* would be useful in preventing infection of the seronegative pregnant woman which would eliminate congenital toxoplasmosis. A vaccine to protect immunocompromised individuals is a possibility, but due to the immune status of these individuals, it does not appear to be a promising approach. At the present time, however, a vaccine to prevent infection by *T. gondii* is not available.

REFERENCES

1. Al-Khalidi, N. W., and J. P. Dubey. 1979. Prevalence of *Toxoplasma gondii* in horses. J. Parasitol. 65:331-334.
2. Al-Khalidi, N. W., S. E. Weisbrode, and J. P. Dubey. 1980. Pathogenicity of *Toxoplasma gondii* oocysts to ponies. Am. J. Vet. Res. 41:1549-1551.
3. August, J. R., and T. M. Chase. 1987. Toxoplasmosis. Vet. Clinics N. Am. Small Anim. Pract. 17:55-71.
4. Barbier, D., T. Ancelle, and G. Martin-Bouyer. 1983. Seroepidemiological survey of toxoplasmosis in La Guadeloupe, French West Indies. Am. J. Trop. Med. Hyg. 32:935-942.
5. Benenson, M. W., E. T. Takafuji, S. M. Lemon, and A. J. Sulzer. 1982. Oocyst-transmitted toxoplasmosis associated with ingestion of contaminated water. N. Engl. J. Med. 307:666-669.
6. Biancifiori, F., C. Rondini, V. Grelloni, and T. Frescura. 1986. Avian toxoplasmosis: experimental infection of chicken and pigeon. Comp. Immun. Microbiol. Infect. Dis. 9:337-346.
7. Brake, R. J., K. D. Murrell, E. E. Ray, J. D. Thomas, B. A. Muggenburg, and J. S. Sivinski. 1985. Destruction of *Trichinella spiralis* by low-dose irradiation of infected pork. J. Food Safety 7:127-143.
8. Braveny, I., H. Janssen, and R. Disko. 1977. Zur Beduetung von Rohfleisch und Katzen als Infektionsquellen der Toxoplalsmose. Bundesgesundheitsblatt 20 (18):259-260.
9. Burridge, M. J., W. J. Bigler, D. J. Forrester, and J. M. Hennemann. 1979. Serologic survey for *Toxoplasma gondii* in wild animals in Florida. J. Am. Vet. Med. Assoc. 175:964-967.
10. Centers for Disease Control. 1975. Toxoplasmosis--Pennsylvania. Morbid. Mortal. Weekly Rep. 24 (34):285-286.
11. Chinchilla, M., and A. Ruiz. 1976. Cockroaches as possible transport hosts of *Toxoplasma gondii* in Costa Rica. J. Parasitol. 62:140-142.
12. Collins, G. H. 1981. Studies in *Sarcocystis* species. VIII. *Sarcocystis* and *Toxoplasma* in red deer (*Cervus elaphus*). N. Zeal. Vet. J. 29:126-127.
13. Costa, A. J., F. G. Araujo, J. O. Costa, J. D. Lima, and E. Nascimento. 1977. Experimental infection of bovines with oocysts of *Toxoplasma gondii*. J. Parasitol. 63:212-218.
14. Desmonts, G. 1962. Epidemiologie de la toxoplasmose. Revue d'Hygiene et de Medicine Sociale 10:201-217.
15. Desmonts, G., J. Couvreur, F. Alison, J. Baudelot, J. Gerbeaux, and M. Lelong. 1965. Etude epidemiologique sur la toxoplasmose: de l'influence de la cuisson des viandes de boucherie sur la frequence de l'infection humaine. Rev. Franc. Etudes Clin. et Biol. 10:952-958.
16. Dubey, J. P. 1980. Persistence of encysted *Toxoplasma gondii* in caprine livers and public health significance of toxoplasmosis in goats. J. Am. Vet. Med. Assoc. 177:1203-1207.
17. Dubey, J. P. 1981. Epizootic toxoplasmosis associated with abortion in dairy goats in Montana. J. Am. Vet. Med. Assoc. 178:661-670.

18. Dubey, J. P. 1981. Isolation of encysted *Toxoplasma gondii* from musculature of moose and pronghorn in Montana. Am. J. Vet. Res. 42:126-127.
19. Dubey, J. P. 1981. *Toxoplasma*-induced abortion in dairy goats. J. Am. Vet. Med. Assoc. 178:671-674.
20. Dubey, J. P. 1982. Isolation of encysted *Toxoplasma gondii* from muscles of mule deer in Montana. J. Am. Vet. Med. Assoc. 181:1535.
21. Dubey, J. P. 1982. Repeat transplacental transfer of *Toxoplasma gondii* in dairy goats. J. Am. Vet. Med. Assoc. 180:1220-1221.
22. Dubey, J. P. 1983. Distribution of cysts and tachyzoites in calves and pregnant cows inoculated with *Toxoplasma gondii* oocysts. Vet. Parasitol. 13:199-211.
23. Dubey, J. P. 1984. Experimental toxoplasmosis in sheep fed *Toxoplasma gondii* oocysts. Int. Goat Sheep Res. 2:93-104.
24. Dubey, J. P. 1985. Persistence of encysted *Toxoplasma gondii* in tissues of equids fed oocysts. Am. J. Vet. Res. 46:1753-1754.
25. Dubey, J. P. 1985. Serologic prevalence of toxoplasmosis in cattle, sheep, goats, pigs, bison, and elk in Montana. J. Am. Vet. Med. Assoc. 186:969-970.
26. Dubey, J. P. 1986. A review of toxoplasmosis in cattle. Vet. Parasitol. 22:177-202.
27. Dubey, J. P. 1986. A review of toxoplasmosis in pigs. Vet. Parasitol. 19:181-223.
28. Dubey, J. P. 1988. Long-term persistence of *Toxoplasma gondii* in tissues of pigs inoculated with *T. gondii* oocyst and effect of freezing on viability of tissue cysts in pork. Am. J. Vet. Res. 49:910-913.
29. Dubey, J. P. 1990. Status of toxoplasmosis in cattle in the United States. J. Am. Vet. Med. Assoc. 196:257-259.
30. Dubey, J. P. 1990. Status of toxoplasmosis in pigs in the United States. J. Am. Vet. Med. Assoc. 196:270-274.
31. Dubey, J. P. 1990. Status of toxoplasmosis in sheep and goats in the United States. J. Am. Vet. Med. Assoc. 196:259-262.
32. Dubey, J. P., and D. S. Adams. 1990. Prevalence of *Toxoplasma gondii* antibodies in dairy goats from 1982 to 1984. J. Am. Vet. Med. Assoc. 196:295-296.
33. Dubey, J. P., and C. P. Beattie. 1988. Toxoplasmosis of animals and man. CRC Press, Inc., Boca Raton, FL.
34. Dubey, J. P., and C. A. Kirkbride. 1989. Economic and public health considerations of congenital toxoplasmosis in lambs. J. Am. Vet. Med. Assoc. 195:1715-1716.
35. Dubey, J. P., and C. A. Kirkbride. 1989. Enzootic toxoplasmosis in sheep in north-central United States. J. Parasitol. 75:673-676.
36. Dubey, J. P., and S. P. Sharma. 1980. Parasitemia and tissue infection in sheep fed *Toxoplasma gondii* cysts. J. Parasitol. 66:111-114.
37. Dubey, J. P., and R. H. Streitel. 1976. Prevalence of *Toxoplasma* infection in cattle slaughtered at an Ohio abattoir. J. Am. Vet. Med. Assoc. 169:1197-1199.
38. Dubey, J. P., N. L. Miller, and J. K. Frenkel. 1970. Characterization of the new fecal form of *Toxoplasma gondii*. J. Parasitol. 56:447-456.
39. Dubey, J. P., K. D. Murrell, and R. Fayer. 1984. Persistence of encysted *Toxoplasma gondii* in tissues of pigs fed oocysts. Am. J. Vet. Res. 45:1941-1943.
40. Dubey, J. P., E. T. Thorne, and S. P. Sharma. 1980. Experimental toxoplasmosis in elk (*Cervus canadensis*). Am. J. Vet. Res. 41:792-793.
41. Dubey, J. P., E. T. Thorne, and E. S. Williams. 1982. Induced toxoplasmosis in pronghorns and mule deer. J. Am. Vet. Med. Assoc. 181:1263-1267.
42. Dubey, J. P., R. J. Brake, K. D. Murrell, and R. Fayer. 1986. Effect of irradiation on the viability of *Toxoplasma gondii* cysts in tissues of mice and pigs. Am. J. Vet. Res. 47:518-522.
43. Dubey, J. P., K. D. Murrell, R. Fayer, and G. A. Schad. 1986. Distribution of *Toxoplasma gondii* cysts in commercial cuts of pork. J. Am. Vet. Med. Assoc. 188:1035-1037.
44. Dubey, J. P., A. W. Kotula, A. Sharar, C. D. Andrews, and D. S. Lindsay. 1990. Effect of high temperature on infectivity of *Toxoplasma gondii* tissue cysts in pork. J. Parasitol. 76:201-204.
45. Dubey, J. P., S. Miller, G. Desmonts, P. Thulliez, and W. R. Anderson. 1986. *Toxoplasma gondii*-induced abortion in dairy goats. J. Am. Vet Med. Assoc. 188:159-162.
46. Dubey, J. P., S. P. Sharma, D. D. Juranek, A. J. Sulzer, and S. M. Teutsch. 1981. Characterization of *Toxoplasma gondii* isolates from an outbreak of toxoplasmosis in Atlanta, Georgia. Am. J. Vet. Res. 42:1007-1010.
47. Dubey, J. P., J. C. Leighty, V. C. Beal, W. R. Anderson, C. D. Andrews, and P. Thulliez. 1991. National seroprevalence of *Toxoplasma gondii* in pigs. J. Parasitol. 77:517-521.
48. Dubey, J. P., S. P. Sharma, C. W. G. Lopes, J. F Williams, C. S. F. Williams, and S. E. Weisbode. 1980. Caprine toxoplasmosis: abortion, clinical signs, and distribution of *Toxoplasma* in tissues of goats fed *Toxoplasma gondii* oocysts. Am. J. Vet. Res. 41:1072-1076.
49. Durfee, P. T., and J. Chien. 1971. Transmission of *Toxoplasma gondii* to cats via ingestion of infected pork. J. Am. Vet. Med. Res. Assoc. 159:1783-1788.
50. Entzeroth, R., G. Piekarski, and E. Scholtyseck. 1981. Fine structural study of a *Toxoplasma* strain from the roe deer. Zeitsch. Parasitenkd. 66:109-112.
51. Faull, W. B., M. J. Clarkson, and A. C. Winter. 1986. Toxoplasmosis in a flock of sheep: some investigations into its source and control. Vet. Rec. 119:491-493.
52. Fayer, R., and J. K. Frenkel. 1979. Comparative infectivity for calves of oocysts of feline coccidia: *Besnoita, Hammondia, Cystoisospora, Sarcocystis*, and *Toxoplasma*. J. Parasitol. 65:756-762.
53. Fertig, A., S. Selwyn, and M. J. K. Tibble. 1977. Tetracycline treatment in a food-borne outbreak of toxoplasmosis. Br. Med. J. 1(6068):1064.
54. Fleck, D. G., B. S. Chessum, and M. Perkins. 1972. Coccidian-like nature of *Toxoplasma gondii*. Br. Med. J. 3:111-112.
55. Franti, C. E., G. E. Connolly, H. P. Riemann, D. E. Behymer, R. Ruppanner, C. M. Willadsen, and W. Longhurst. 1975. A survey for *Toxoplasma gondii* antibodies in deer and other wildlife on a sheep range. J. Am. Vet. Med. Assoc. 167:565-568.
56. Frenkel, J. K. 1990. Toxoplasmosis in human beings. J. Am. Vet. Med. Assoc. 196:240-248.
57. Frenkel, J. K., and J. P. Dubey. 1972. Toxoplasmosis and its prevention in cats and man. J. Infect. Dis. 126:664-673.
58. Frenkel, J. K., and A. Ruiz. 1981. Endemicity of toxoplasmosis in Costa Rica. Transmission between cats, soil, intermediate hosts, and humans. Am. J. Epidemiol. 113:254-269.
59. Frenkel, J. K., J. P. Dubey, and N. L. Miller. 1970. *Toxoplasma gondii* in cats: fecal stages identified as coccidian oocysts. Science 167:893-896.
60. Frenkel, J. K., A. Ruiz, and M. Chinchilla. 1975. Soil survival of *Toxoplasma* oocysts in Kansas and Costa Rica. Am. J. Trop. Med. Hyg. 24:439-443.
61. Gellin, B. G., and R. Soave. 1992. Coccidian infections in AIDS. Med. Clinics of North America 76:205-234.
62. Ghorbani, M., G. E. Edrissian, and N. Assad. 1978. Serological survey of toxoplasmosis in the northern part of Iran, using indirect fluorescent antibody technique. Trans. Roy. Soc. Trop. Med. Hyg. 72:369-371.
63. Hermentin, K., and H. Aspock. 1988. Efforts towards a vaccine against Toxoplasma gondii: a review. Zbl. Bakt. Hyg. A 269:423-436.
64. Hollins, P. J., B. I. Hoffbrand, and I. M. Haffajee. 1972. Toxoplasma encephalitis in a raw steak eater. Postgrad. Med. J. 48:384-385.
65. Huskinson-Mark, J., F. G. Araujo, and J. S. Remington. 1991. Evaluation of the effects of drugs on the cyst form of *Toxoplasma gondii*. J. Infect. Dis. 164:170-177.
66. Hutchison, W. M., J. F. Dunachie, J. C. Siim, and K. Work. 1970. Coccidian-like nature of *Toxoplasma gondii*. Brit. Med. J. 1970, I, 142-144.
67. Jackson, M. H., and W. M. Hutchinson. 1989. The prevalence and source of *Toxoplasma* infection in the environment. Adv. Parasitol. 28:55-105.
68. Jacobs, L. 1963. *Toxoplasma* and toxoplasmosis. Annu. Rev. Microbiol. 17:429-450.
69. Jacobs, L., and M. L. Melton. 1966. Toxoplasmosis in chickens. J. Parasitol. 52:1158-1162.
70. Jacobs, L., J. S. Remington, and M. L. Melton. 1960. A survey of meat samples from swine, cattle, and sheep for the presence of encysted *Toxoplasma*. J. Parasitol. 46:23-28.
71. Jacobs, L., J. S. Remington, and M. L. Melton. 1960. The resistance of the encysted form of *Toxoplasma gondii*. J. Parasitol. 46:11-21.
72. Kean, B. H., A. C. Kimball, and W. N. Christenson. 1969. An epidemic of acute toxoplasmosis. J. Am. Med. Assoc. 208:1002-1004.
73. Knaus, B. U. 1975. Untersuchungen zur Bedeutung von Rohfleischgenuss und Tierkontakt fur die Toxoplasma-Infektion des Menschen. Z. Gesamte Hyg. Ihre Grenzgeb. 21:61-64.

74. Konishi, E., and J. Takahashi. 1987. Some epidemiological aspects of *Toxoplasma* infections in a population of farmers in Japan. Int. J. Epidemiol. 16:277-281.
75. Kotula, A. W., K. D. Murrell, L. Acosta-Stein, L. Lamb, and L. Douglass. 1983. *Trichinella spiralis*: effect of high temperature on infectivity in pork. Exp. Parasitol. 56:15-19.
76. Kotula, A. W., J. P. Dubey, A. K. Sharar, C. D. Andrews, S. K. Shen, and D. S. Lindsay. 1991. Effect of freezing on infectivity of *Toxoplasma gondii* tissue cysts in pork. J. Food Prot. 54:687-690.
77. Kotula, A. W., A. K. Sharar, E. Paroczay, H. R. Gamble, K. D. Murrell, and L. Douglass. 1990. Infectivity of *Trichinella spiralis* from frozen pork. J. Food Prot. 53:571-573.
78. Leighty, J. C. 1990. Strategies for control of toxoplasmosis. J. Am. Vet. Med. Assoc. 196:281-286.
79. Lindsay, D. S., B. L. Blagburn, J. P. Dubey, and W. H. Mason. 1991. Prevalence and isolation of *Toxoplasma gondii* from white-tailed deer in Alabama. J. Parasitol. 77:62-64.
80. Luft, B. J., and J. S. Remington. 1984. Acute *Toxoplasma* infection among family members of patients with acute lymphadenopathic toxoplasmosis. Arch. Intern. Med. 144:53-56.
81. Lunden, A., and A. Uggla. 1992. Infectivity of *Toxoplasma gondii* in mutton following curing, smoking, freezing or microwave cooking. Int. J. Food Microbiol. 15:357-363.
82. McDonald, J. C., T. W. Gyorkos, B. Alberton, J. D. MacLean, G. Richer, and D. Juranek. 1990. An outbreak of toxoplasmosis in pregnant women in northern Quebec. J. Infect. Dis. 161:769-774.
83. Magaldi, C., H. Elkis, D. Pattoli, and A. L. Coscina. 1969. Epidemic of toxoplasmosis at a university in Sao-Jose-dos-Campos, S.P.—Brazil. 1. Clinical and serologic data. Rev. Lat-Am. Microbiol. Parasitol. 11:5-13.
84. Masur, H., T. C. Jones, J. A. Lempert, and T. D. Cherubini. 1978. Outbreak of toxoplasmosis in a family and documentation of acquired retinochoroiditis. Am. J. Med. 64:396-402.
85. Miller, N. L., J. K. Frenkel, and J. P. Dubey. 1972. Oral infections with toxoplasma cysts and oocysts in felines, other mammals, and birds. J. Parasitol. 58:923-937.
86. O'Donoghue, P. J., M. J. Riley, and J. F. Clarke. 1987. Serological survey for *Toxoplasma* infections in sheep. Aust. Vet. J. 64:40-45.
87. Prickett, M. D., M. W. Dreesen, W. D. Waltman, J. L. Blue, and J. Brown. 1985. Correlation of tissue infection and serologic findings in pigs fed *Toxoplasma gondii* oocysts. Am. J. Vet. Res. 46:1130-1132.
88. Remington, J. S., B. Efron, E. Cavanaugh, H. J. Simon, and A. Trejos. 1970. Studies on toxoplasmosis in El Salvador. Prevalence and incidence of toxoplasmosis as measured by the Sabin-Feldman dye test. Trans. Roy. Soc. Trop. Med. Hyg. 64:252-267.
89. Riemann, H. P., M. E. Meyer, J. H. Theis, G. Kelso, and D. E. Behymer. 1975. Toxoplasmosis in an infant fed unpasteurized goat milk. J. Pediatr. 87:573-576.
90. Riemann, H. P., C. M. Willadsen, L. J. Berry, D. E. Behmeyer, Z. V. Garcia, C. E. Franti, and R. Ruppanner. 1977. Survey for toxoplasma antibodies among sheep in the western United States. J. Am. Vet. Med. Assoc. 171:1260-1264.
91. Riemann, H. P., A. T. Smith, C. Stormont, R. Ruppanner, D. E. Behymer, Y. Suzuki, C. E. Franti, and B. B. Verma. 1975. Equine toxoplasmosis: a survey for antibodies to *Toxoplasma gondii* in horses. Am. J. Vet. Res. 36:1797-1800.
92. Roher, D. P., M. J. Ryan, S. W. Nielsen, and D. E . Roscoe. 1981. Acute fatal toxoplasmosis in squirrels. J. Am. Vet. Med. Assoc. 179:1099-1101.
93. Rothe, J., P. J. McDonald, and A. M. Johnson. 1985. Detection of *Toxoplasma* cysts and oocysts in an urban environment in a developed country. Pathology 17:497-499.
94. Ruiz, A., J. K. Frenkel, and L. Cerdas. 1973. Isolation of Toxoplasma from soil. J. Parasitol. 59:204-206.
95. Sacks, J. J., R. R. Roberto, and N. F. Brooks. 1982. Toxoplasmosis infection associated with raw goat's milk. J. Am. Med. Assoc. 248:1728-1732.
96. Sacks, J. J., D. G. Delgado, H. O. Lobel, and R. L. Parker. 1983. Toxoplasmosis infection associated with eating undercooked venison. Am. J. Epidemiol. 118:832-838.
97. Sharma, S. P., and J. P. Dubey. 1981. Quantitative survival of *Toxoplasma gondii* tachyzoites and bradyzoites in pepsin and trypsin solutions. Am. J. Vet. Res. 42:128-130.
98. Sheffield, H. G., and M. L. Melton. 1970. *Toxoplasma gondii*: the oocyst, sporozoite, and infection of cultured cells. Science 167:892-893.
99. Shen, L., L. Zhichung, Z. Biaucheng, and Y. Huayuan. 1990. Prevalence of *Toxoplasma gondii* infection in man and animals in Guangdong, Peoples Republic of China. Vet. Parasitol. 34:357-360.
100. Silva, L. M. de, D. L. Mulcahy, and K. R. Kamath. 1984. A family outbreak of toxoplasmosis: a serendipitous finding. J. Infect. 8:163-167.
101. Skinner, L. J., A. C. Timperley, D. Wightman, A. W. Chatterton, and D. O. Ho-Yen. 1990. Simultaneous diagnosis of toxoplasmosis in goats and goatowner's family. Scand. J. Infect. Dis. 22:359-361.
102. Sousa, O. E., R. E. Saenz, and J. K. Frenkel. 1988. Toxoplasmosis in Panama: a 10-year study. Am. J. Trop. Med. Hyg. 38:315-322.
103. Stagno, S., A. C. Dykes, C. S. Amos, R. A. Head, D. D. Juranek, and K. Walls. 1980. An outbreak of toxoplasmosis linked to cats. Pediatrics 65:706-712.
104. Stalheim, O. H., W. T. Hubbert, A. D. Boothe, W. J. Zimmermann, D. E. Hughes, D. Barnett, J. L. Riley, and J. Foley. 1980. Experimental toxoplasmosis in calves and pregnant cows. Am. J. Vet. Res. 41:10-13.
105. Teutsch, S. M., D. D. Juranek, A. Sulzer, J. P. Dubey, and R. K. Sikes. 1979. Epidemic toxoplasmosis associated with infected cats. N. Engl. J. Med. 300:695-699.
106. Todd, E. C. D. 1989. Preliminary estimates of costs of foodborne disease in the United States. J. Food Prot. 52:595-601.
107. Ven, E. van de, W. Melchers, J. Galama, W. Camps, and J. Meusissen. 1991. Identification of *Toxoplasma gondii* infections by BI gene amplification. J. Clin. Microbiol. 29:2120-2124.
108. Waldeland, H. 1976. Toxoplasmosis in sheep. Toxoplasma gondii in muscular tissue, with particular reference to dye test titres and haemoglobin type. Acta Vet. Scand. 17:403-411.
109. Wallace, G. D. 1971. Experimental transmission of *Toxoplasma gondii* by filth-flies. Am. J. Trop. Med. Hyg. 20:411-413.
110. Wallace, G. D. 1972. Experimental transmission of *Toxoplasma gondii* by cockroaches. J. Infect. Dis. 126:545-547.
111. Wallace, G. D. 1976. The prevalence of toxoplasmosis on Pacific Islands and the influence of ethnic group. Am. J. Trop. Med. Hyg. 25:48-53.
112. Weinman, D. 1952. Toxoplasma and toxoplasmosis. Annu. Rev. Microbiol. 6:281-298.
113. Weinman, D., and A. H. Chandler. 1956. Toxoplasmosis in man and swine—an investigation of the possible relationship. J. Am. Med. Assoc. 161:229-232.
114. Williamson, J. M. W., H. Williams, and G. A. M. Sharman. 1980. Toxoplasmosis in farmed red deer (*Cervus elaphus*) in Scotland. Res. Vet. Sci. 29:36-40.
115. Work, K. 1971. Toxoplasmosis. With special reference to transmission and life cycle of *Toxoplasma gondii*. Acta Path. Microbiol. Scand., Sect. B 1971, Supple. No. 221.
116. Yilmaz, S. M., and S. H. Hopkins. 1972. Effects of different conditions on duration of infectivity of *Toxoplasma gondii* oocysts. J . Parasitol. 58:938-939.